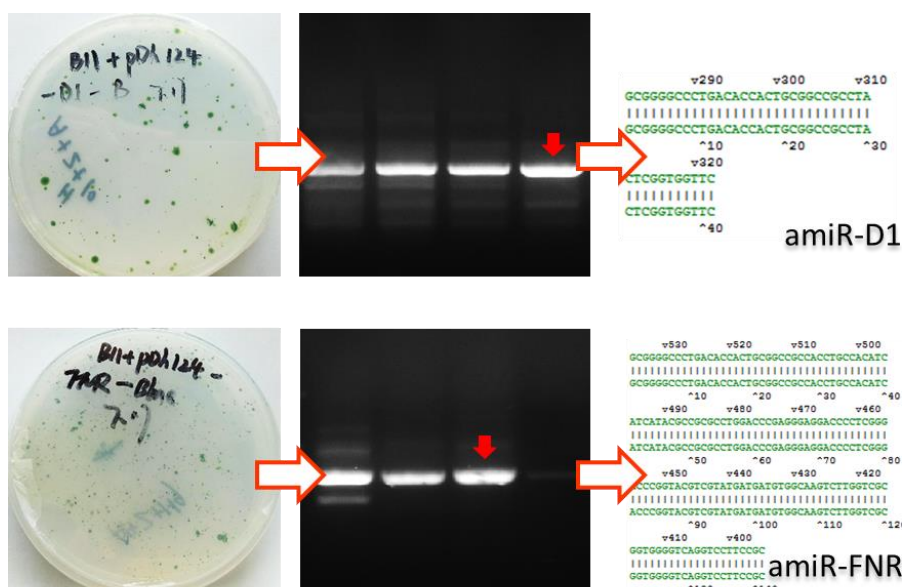


## Result

*First, the selection of gene-modified green algae's positive transformants.*

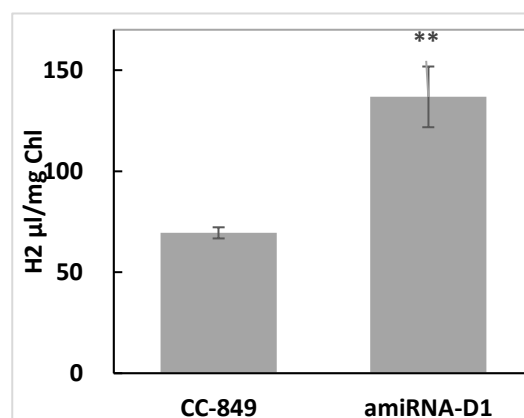
We well-distributed the gene-modified algae strains transformed by two vectors (pDb12-BD-CIB1 and pDh124-VP16-CRY2-UAS-miRNA) on the double resistant TAP solid medium plate that included paromycin and hygromycin B. After the “green” on the plate was all disappeared, the new monoclonal

*algae were the selected transformants. Then the gene-modified algae were enlarged cultivated, from which we extracted the genome DNA. We used amiRNA's precursor primer to have the DNA proceed polymerase chain reaction. The glue with correct band size was cut and was examined by sequencing. The above pictures show amiR-D1's transformation and selection, the below pictures show amiR-FNR's transformation and selection.*



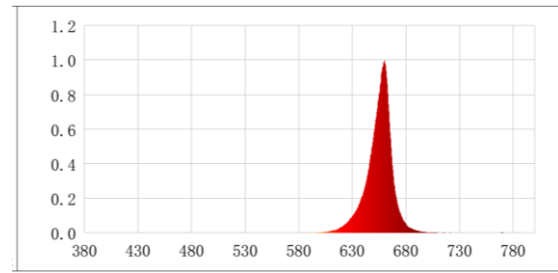
Second, the Hydrogen yield detection--  
--- measured by gas chromatography

1. We cultivated the gene-modified green algae in blue light (The wave length is about 450 nm) and induced the expression of relevant miRNA. When they reached logarithm stage, we measured hydrogen production. The experiment results indicated that the hydrogen production of the transformants increased after induced with blue light. The result



*proofed that our designed microRNA did have effect on inhabiting the photosynthesis of green algae reaction system.*

2. We also cultivated the gene-modified green algae in the condition lacking in blue light. ( Red light LED plant growth lamp, peak wave length: 660nm, half width: 17.9nm, dominant wave length: 642.4 nm, purity: 0.989).



When the green algae reached logarithm stage, we used blue light, whose wave length is about 450 nm, to induce the expression of the relevant miRNA and increase hydrogen production. First we used blue light to induce the expression of miRNA and measured hydrogen production every 4 hours. After 8 hours, we re-cultivated the green algae in red light. After 16 hours, again we used blue light to induce the expression of miRNA and measured hydrogen production every 4 hours for 12 hours. The results indicates that the hydrogen production of will increase when induced with blue light. After re-cultivated with red light and repeating induced with blue light, the hydrogen production will increase again. This result proofs that it's effective for the light-mediated expression system. If repeating to induce the green algae with blue light, we can ultimately make this system produce hydrogen continuously.

